Figure 1

Figure 2

Figure 3

Figure 4A

```
                FR1                                CDR1         FR2              CDR2                       FR3                                        CDR3                   FR4
        ---------------------------------         -------    -------------    -------------------    --------------------------------------     ----------------------     -----------
                 1         2         3                 1ab2345        4               6                    7         8         9                         1                  1 1
        1234567890123456789012345670             1ab2345    6789012345456789  012abc3456789012345   67890123456789012abc345678901234           567890abcdefghijklm12      0 1
                                                                                                                                                                            34567890123
VH EL-14 QVQLVQSGAEAKKPGSSVKVSCKASGDTFN           S--FPIS    WVRQAPGQGGLEWMG   GIIP--IFGSTKYAQKFQG    RVTMTADGSTSTAYMELNSLRSEDTAIYYCAR            QQNGGWYEGPLLEPRPDALDI     WGQGTMVTVSS
DP-10    ---------------V--------G--S             ---YA--    ----------------  -------TAN---------    ----I--E-------S-------V--------
```

```
                FR1                                CDR1         FR2              CDR2                       FR3                                        CDR3                     FR4
        ---------------------------------         -------    -------------    -------------------    --------------------------------------     ------------------------     -----------
                 1         2         3                 1ab2345        4               5                    7         8         9                         1                    1 1
        12345678901234567891234567890            1ab2345    67890123456789    012abc3456789012345   67890123456789012abc345678901234           567890abcdefghijklmno12      0 1
                                                                                                                                                                              34567890123
VH IT-2  QVQLLQSATEVKKPGASMKVSCMASGYPFT           S--YDIS    WVRQAPGQGGLEWMG   WISI--YSGNTDYAQKFQG    RVTMTTDTSRRTAYMELRSLRSDDTAVYYCAR            DGGGGAYEDVWSGEYPEYYAMDV      WGQGTTVTVSS
DP-14    ----GA--------V----K----T--              ----G--    ---------------   ----A--N--N----L---    --------------------TS----------
```

```
           FR1                    CDR1      FR2          CDR2              FR3                              CDR3                     FR4
         --------                -------   --------    --------    -------------------------------    ----------------------    ------------
              1         2    3             4             5     6          7        8        9              1                      1    1
         1234567890123456789012345678890  1ab2345   67890123456789  012abc3456789012345  6789012345678901234abc34567890123 4  567890abcdefghijklmno12  34567890123

DP-14    QVQLVQSGAEVKKPGASVKVSCKASGYTFT    S--YGIS   WVRQAPGQGLEWMG  WISA--YNGNTNYAQKLQG  RVTMTTDTSTSTAYMELRSLRSDDTAVYYCAR   DGGGGAYEDVWSGEYPEYYAMDV  WGQGTTVTVSS
VH IT-2  -----L-AT-----M----M---P--        ----D--   -------------   ----I---S--D---F--   ------RR-------------
VH EL-5  -----L-AT-----M----M---P--        ----D--   -------------V--  ------H---F--      -------RR-------------
VH EL-25 -----L--A---R----------P--        ----D--   ------------i--  ----I---S--D---F--  ------RR-------------
```

Figure 4B

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Figure 4C

CDR3

EL-14   QQNGGWYEGPLLEPRPD--ALDI
         || || |. |.|. .|.
IT-2   DGGGGAYEDVWSGEYPEYYAMDV

Figure 5

Figure 6

Figure 7A NEUTRALIZATION BY scFv EL-14

Figure 7B NEUTRALIZATION BY scFv IT-2

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Figure 8

A — Selection IT-FVIII

B — Selection CLB-CAg 117-80K

Legend:
- CAg 12-80K
- CAg 12-A3C1
- CAg 12

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Deduced protein sequences of isolated FVIII A3-C1 specific scFv

Heavy chains

```
                FR1              CDR1    FR2       CDR2          FR3                                                    CDR3                              FR4

                                                                                                                       1         1
                                                           6                  8         9                    0        0         1
         1234567890123456789012345 67890 12345 678901234567890 012a345678901234 56789012345678901234 567890abcdefghil2 34567890123

DP-15    QVQLVQSGAEVKKPGASVKVSCKASGYTFT SYDIN WVRQATGQGLEWMG WMNPNSGNTGYAQKFQG RVTMTRNTSISTAYMELSSLRSEDTAVYYCAR ----- CDTILLIWFGPAPYNDS WGQGTLV
B38      ---L-YA-D----------T---I-----    ------------------ --A-F----K- --L-L--D--T------ -----RN-E--------

DP-31    EVQLVESGGGLVQPGRSLRLSCAASGFTFD DYAMH WVRQAPGKGLEWVS GISWNSGSIGYADSVKG RFTISRDNAKNSLYLQMNSLRAEDTAILYYCAK D
B18      Q----Q----K-----------------I-    -----E------- --VT-SGTT--F----- -----Y--------   -----L PYINSSNYRRGVAAFDI WGGGTMVTVSS

DP-49    QVQLVESGGGVVQPGRSLRLSCAASGFTFS SYGMH WVRQAPGKGLEWVA VISYDGSNKYYADSVKG RFTISRDNSKNTLYLQMNSLRAEDTAVYYCAK
B35      E---------L------------VD-L--    -------A----- ------ND--------- --A---A------TI--            DLIESNI            AEAL WGQGTLVTVSS

DP-77    EVQLVESGGGLVKPGGSLRLSCAASGFTFS SYSMN WVRQAPGKGLEWVS SISSSSSYIYYADSVKG RFTISRDNAKNSLYLQMNSLRAEDTAVYYCAR
B04      ----K--E----------------R R-DIH --T------ --GGN--D------   ------N-VV---M--F--- DGTIFGSAATWR AFDI WGRGTMVTVSSG
```

Figure 9A

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```
+1    Gln  Val  Gln  Leu  Leu  Gln  Ser  Ala  Ala  Asp   Val  Lys  Lys  Pro  Gly  Ala  Ser
      CAGGTGCAGC TGTTGCAGTC TGCAGCTGAC GTGAAGAAGC CTGGGGCCTC 50
      GTCCACGTCG ACAACGTCAG ACGTCGACTG CACTTCTTCG GACCCCGGAG

+1    Val  Lys  Val  Ser  Cys  Thr  Ala  Ser  Gly  Tyr  Ile  Phe  Thr  Ser  Tyr  Asp  Ile
      AGTGAAGGTC TCCTGTACGG CTTCTGGATA CATCTTCACC AGTTATGATA 100
      TCACTTCCAG AGGACATGCC GAAGACCTAT GTAGAAGTGG TCAATACTAT

+1    Asn  Trp  Val  Arg  Gln  Ala  Thr  Gly  Gln  Gly  Leu  Glu  Trp  Met  Gly  Trp
      TCAACTGGGT GCGACAGGCC ACTGGACAAG GGCTTGAGTG GATGGGATGG 150
      AGTTGACCCA CGCTGTCCGG TGACCTGTTC CCGAACTCAC CTACCCTACC

+1    Met  Asn  Pro  Asn  Ser  Gly  Asn  Ala  Gly  Phe  Ala  Gln  Lys  Phe  Lys  Gly  Arg
      ATGAATCCTA ACAGTGGTAA CGCAGGCTTT GCACAGAAGT TTAAGGGCAG 200
      TACTTAGGAT TGTCACCATT GCGTCCGAAA CGTGTCTTCA AATTCCCGTC

+1    Leu  Thr  Leu  Thr  Arg  Asp  Thr  Ser  Thr  Ser  Thr  Ala  Tyr  Met  Glu  Leu  Arg
      ACTCACCTTG ACCAGGGACA CTTCCACAAG CACAGCCTAC ATGGAGCTGA 250
      TGAGTGGAAC TGGTCCCTGT GAAGGTGTTC GTGTCGGATG TACCTCGACT

+1    Arg  Leu  Glu  Ser  Glu  Asp  Thr  Ala  Val  Tyr  Tyr  Cys  Ala  Arg  Cys  Asp
      GGAGACTGGA ATCTGAGGAC ACGGCCGTGT ATTACTGTGC GAGATGTGAC 300
      CCTCTGACCT TAGACTCCTG TGCCGGCACA TAATGACACG CTCTACACTG

+1    Thr  Thr  Leu  Leu  Ile  Trp  Phe  Gly  Pro  Ala  Pro  Tyr  Tyr  Asp  Ser  Trp  Gly
      ACCACACTCT TAATCTGGTT CGGGCCCGCC CCCTACTATG ACTCCTGGGG 350
      TGGTGTGAGA ATTAGACCAA GCCCGGGCGG GGGATGATAC TGAGGACCCC

+1    Gln  Gly  Thr  Leu  Val
      CCAGGGAACT CTAGTC                                        400
      GGTCCCTTGA GATCAG
```
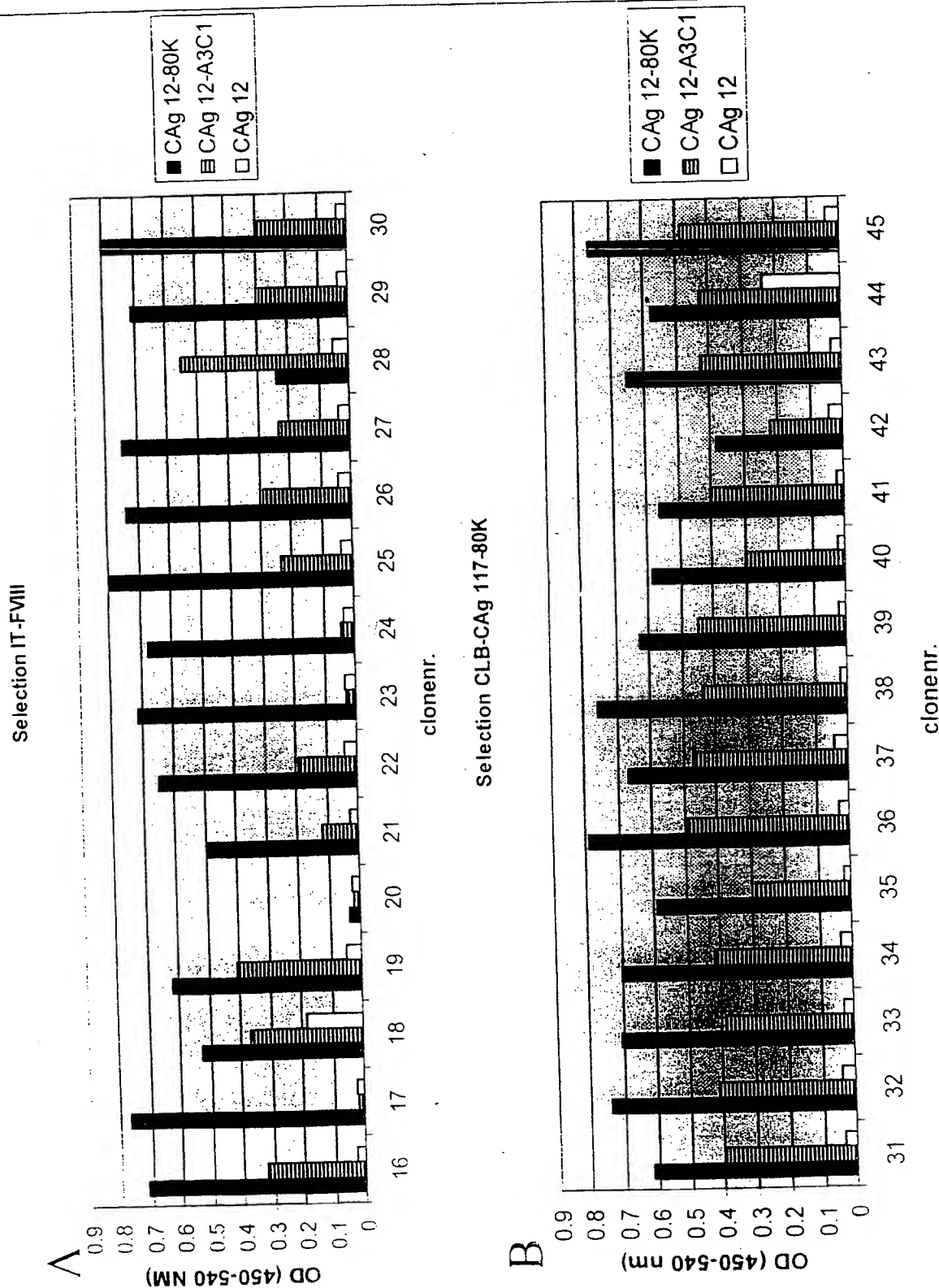
Figure 9B

```
+1    Gln  Val  Gln  Leu  Val  Gln  Ser  Gly  Gly  Gly    Leu  Val  Gln  Pro  Gly  Lys  Ser
      CAGGTGCAAC TGGTGCAGTC TGGGGGAGGC TTGGTACAGC CTGGCAAGTC 50
      GTCCACGTTG ACCACGTCAG ACCCCCTCCG AACCATGTCG GACCGTTCAG

-1    Leu  Arg  Leu    Ser  Cys  Ala  Ala  Ser  Gly  Phe  Thr  Phe  Gly    Asp  Tyr  Ala  Ile
      CCTGAGACTC TCCTGTGCAG CCTCTGGATT CACATTTGGC GATTATGCCA 100
      GGACTCTGAG AGGACACGTC GGAGACCTAA GTGTAAACCG CTAATACGGT

+1    His  Trp  Val  Arg  Gln  Ala    Pro  Gly  Glu  Gly  Leu  Glu  Trp  Val  Ser  Gly
      TACACTGGGT CCGGCAAGCT CCAGGGGAGG GCCTGGAGTG GGTCTCAGGT 150
      ATGTGACCCA GGCCGTTCGA GGTCCCCTCC CGGACCTCAC CCAGAGTCCA

-1    Val  Thr  Trp  Ser  Gly  Thr  Thr  Ile  Gly  Phe  Ala  Asp  Ser  Val  Lys  Gly  Arg
      GTTACTTGGA GTGGTACTAC TATAGGCTTT GCGGACTCTG TGAAGGGCCG 200
      CAATGAACCT CACCATGATG ATATCCGAAA CGCCTGAGAC ACTTCCCGGC

+1    Phe  Thr  Ile  Ser  Arg  Asp  Asn  Ala  Lys  Asn  Ser  Leu  Tyr  Leu  Tyr  Met  Asn
      ATTCACCATC TCCAGAGACA ACGCCAAGAA TTCCCTGTAT CTGTACATGA 250
      TAAGTGGTAG AGGTCTCTGT TGCGGTTCTT AAGGGACATA GACATGTACT

+1    Ser  Leu  Arg  Ala  Glu  Asp    Thr  Ala  Leu  Tyr  Tyr  Cys  Ala  Leu  Pro  Tyr
      ACAGTCTGAG AGCTGAAGAC ACGGCCTTGT ATTATTGTGC CTTACCATAT 300
      TGTCAGACTC TCGACTTCTG TGCCGGAACA TAATAACACG GAATGGTATA

-:    Ile  Asn  Ser  Ser  Asn  Tyr  Arg  Arg  Gly  Val    Ala  Ala  Phe  Asp  Ile  Trp  Gly
      ATCAACTCGT CCAACTACAG AAGAGGGGTC GCTGCCTTCG ATATCTGGGG 350
      TAGTTGAGCA GGTTGATGTC TTCTCCCCAG CGACGGAAGC TATAGACCCC

-:    Gln  Gly  Thr    Met  Val  Thr  Val  Ser  Ser
      CCAAGGGACA ATGGTCACCG TGTCGAGT                          400
      GGTTCCCTGT TACCAGTGGC ACAGCTCA
```

Figure 9C

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```
    ┌Glu┐ ┌Val┐ ┌Gln┐ ┌─Leu─┐ ┌Val┐ ┌Glu┐ ┌─Ser─┐ ┌Gly┐ ┌Gly┐ ┌Gly┐   ┌Leu┐ ┌Val┐ ┌Gln┐ ┌─Pro─┐ ┌Gly┐ ┌Arg┐ ┌─Ser─
+1  GAGGTGCAGC TGGTGGAGTC TGGGGGAGGC TTGGTACAGC CTGGGAGGTC 50
    CTCCACGTCG ACCACCTCAG ACCCCCTCCG AACCATGTCG GACCCTCCAG

    _  ┌Leu┐ ┌Arg┐ ┌Leu┐  ┌Ser┐ ┌Cys┐ ┌Val┐ ┌─Asp─┐  ┌Ser┐ ┌Gly┐ ┌─Leu┐ ┌Thr┐ ┌Phe┐ ┌Ser┐   ┌Ser┐ ┌Tyr┐ ┌Gly┐ Met
+1  CCTGAGACTC TCCTGTGTAG ACTCTGGACT CACCTTCAGT AGTTATGGCA 100
    GGACTCTGAG AGGACACATC TGAGACCTGA GTGGAAGTCA TCAATACCGT

    _  ┌His┐ ┌Trp┐ ┌─Val─┐ ┌Arg┐ ┌Gln┐ ┌Ala┐   ┌Pro┐ ┌Gly┐ ┌Ala┐ ┌─Gly─┐ ┌Leu┐ ┌Glu┐ ┌─Trp─┐ ┌Val┐ ┌Ala┐ ┌Val┐
+1  TGCACTGGGT CCGCCAGGCT CCAGGCGCGG GGCTGGAGTG GGTGGCCGTT 150
    ACGTGACCCA GGCGGTCCGA GGTCCGCGCC CCGACCTCAC CCACCGGCAA

    ┌Ile┐ ┌Ser┐ ┌Tyr┐ ┌─Asp─┐ ┌Gly┐ ┌Asn┐ ┌─Asp─┐ ┌Lys┐ ┌─Tyr─┐ ┌Tyr┐   ┌Ala┐ ┌Asp┐ ┌Ser┐ ┌─Val─┐ ┌Lys┐ ┌Gly┐ ┌Arg─
-1  ATTTCATACG ACGGAAATGA TAAATATTAT GCAGACTCCG TGAAGGGCCG 200
    TAAAGTATGC TGCCTTTACT ATTTATAATA CGTCTGAGGC ACTTCCCGGC

    _  ┌Phe┐ ┌Ala┐ ┌Ile┐  ┌Ser┐ ┌Arg┐ ┌Asp┐ ┌─Asn─┐ ┌Ala┐ ┌Lys┐ ┌─Asn─┐ ┌Thr┐ ┌Leu┐ ┌Tyr┐   ┌Leu┐ ┌Gln┐ ┌Met┐ Asn
+1  ATTCGCCATC TCCAGAGACA ATGCCAAGAA CACGCTGTAT CTGCAAATGA 250
    TAAGCGGTAG AGGTCTCTGT TACGGTTCTT GTGCGACATA GACGTTTACT

    _  ┌Ser┐ ┌Leu┐ ┌─Thr─┐ ┌Ile┐ ┌Glu┐ ┌Asp┐   ┌Thr┐ ┌Ala┐ ┌Val┐ ┌─Tyr─┐ ┌Tyr┐ ┌Cys┐ ┌─Ala─┐ ┌Lys┐ ┌Asp┐ ┌Leu┐
+1  ACAGCCTGAC AATAGAGGAC ACGGCTGTCT ATTATTGTGC GAAAGATCTC 300
    TGTCGGACTG TTATCTCCTG TGCCGACAGA TAATAACACG CTTTCTAGAG

    _  ┌Ile┐ ┌Glu┐ ┌Ser┐ ┌─Asn─┐ ┌Ile┐ ┌Ala┐ ┌─Glu─┐ ┌Ala┐ ┌Leu┐ ┌Trp┐   ┌Gly┐ ┌Gln┐ ┌Gly┐ ┌─Thr─┐ ┌Leu┐ ┌Val┐ ┌─Thr─
+1  ATAGAATCAA ATATTGCGGA GGCCCTCTGG GGCCAGGGAA CCCTGGTCAC 350
    TATCTTAGTT TATAACGCCT CCGGGAGACC CCGGTCCCTT GGGACCAGTG

    _  ┌Val┐ ┌Ser┐ ┌Ser┐                                              400
+1  CGTGTCGAGT
    GCACAGCTCA
```

Figure 9D

+1 Glu Val Gln Leu Val Lys Ser Gly Glu Gly   Leu Val Lys Pro Gly Gly Ser
```
GAGGTGCAGC TGGTGAAGTC TGGGGAAGGC CTGGTCAAGC CTGGGGGGTC 50
CTCCACGTCG ACCACTTCAG ACCCCTTCCG GACCAGTTCG GACCCCCCAG
```

-1   Leu Arg Leu   Ser Cys Ala Ala Ser Gly Phe Thr Phe Arg   Arg Tyr Asp Ile
```
CCTGAGACTC TCCTGTGCAG CCTCTGGATT CACCTTCAGG AGATATGATA 100
GGACTCTGAG AGGACACGTC GGAGACCTAA GTGGAAGTCC TCTATACTAT
```

+1   His Trp Val Arg Gln Thr   Pro Gly Lys Gly Leu Glu Trp Val Ser Ser
```
TCCACTGGGT CCGCCAGACT CCAGGGAAGG GCCTGGAGTG GGTCTCATCC 150
AGGTGACCCA GGCGGTCTGA GGTCCCTTCC CGGACCTCAC CCAGAGTAGG
```

+1 Ile Ser Ser Gly Gly Asn Tyr Ile Asp Tyr   Ala Asp Ser Val Lys Gly Arg
```
ATCAGTAGTG GTGGTAATTA CATAGACTAC GCAGACTCTG TGAAGGGCCG 200
TAGTCATCAC CACCATTAAT GTATCTGATG CGTCTGAGAC ACTTCCCGGC
```

+1   Phe Thr Ile   Ser Arg Asp Asn Ala Asn Asn Val Val Tyr Leu Gln Met Asn
```
ATTCACCATC TCCAGAGACA ACGCCAACAA TGTTGTCTAT CTACAAATGA 250
TAAGTGGTAG AGGTCTCTGT TGCGGTTGTT ACAACAGATA GATGTTTACT
```

+1   Ser Leu Arg Ala Glu Asp Met Ala Val Tyr Phe Cys Ala Arg Asp Gly
```
ACAGCCTGAG AGCCGAGGAC ATGGCTGTCT ATTTCTGTGC GAGAGATGGG 300
TGTCGGACTC TCGGCTCCTG TACCGACAGA TAAAGACACG CTCTCTACCC
```

-1 Thr Ile Phe Gly Ser Ala Ala Thr Trp Arg Ala Phe Asp Ile Trp Gly Arg
```
ACGATTTTTG GATCGGCGGC GACCTGGCGG GCTTTTGATA TCTGGGGCCG 350
TGCTAAAAAC CTAGCCGCCG CTGGACCGCC CGAAAACTAT AGACCCCGGC
```

-1 Gly Thr Met Val Thr Val Ser Ser
```
GGGGACAATG GTCACCGTGT CGAGT          400
CCCCTGTTAC CAGTGGCACA GCTCA
```
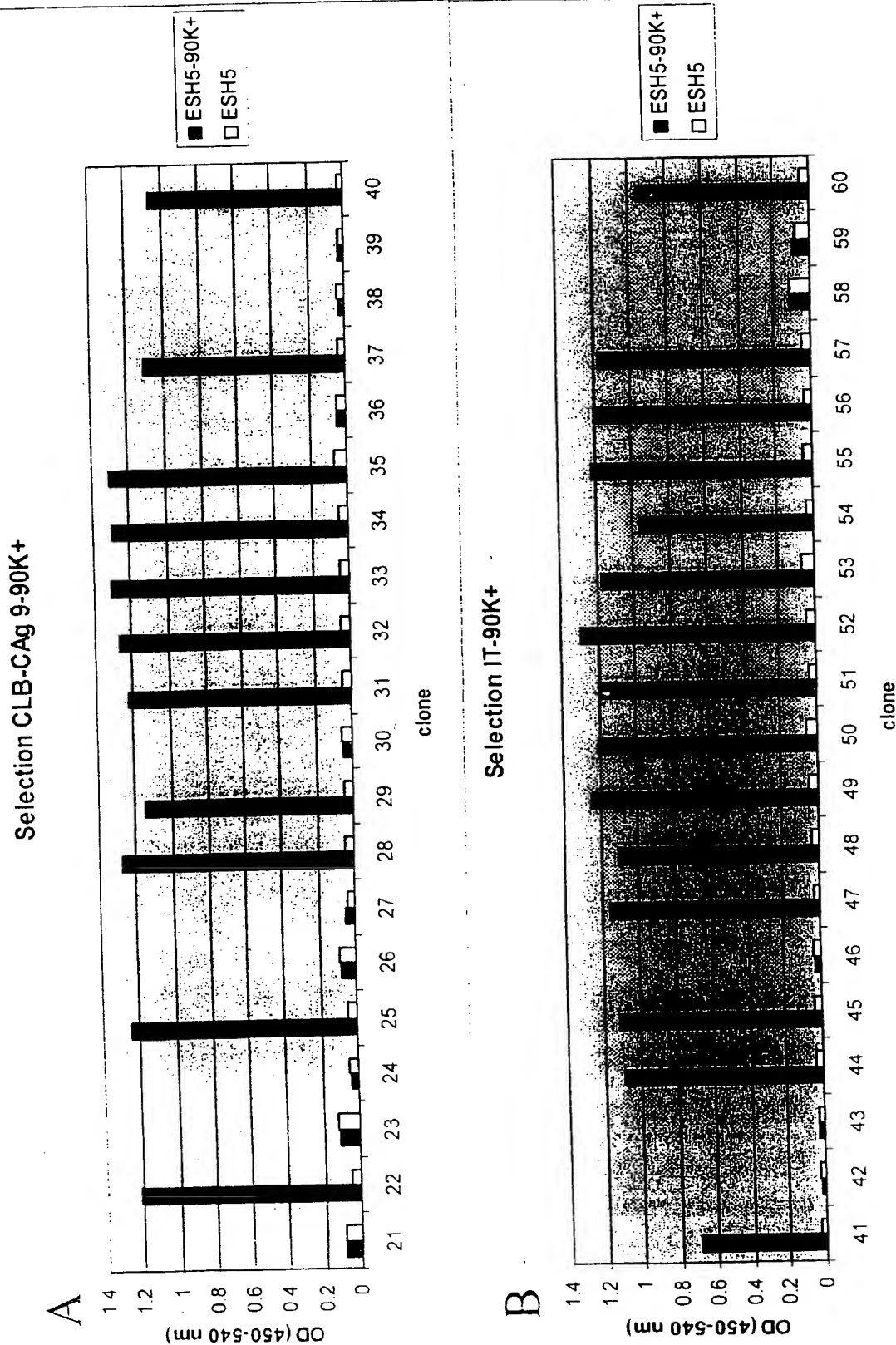
Figure 9E

Figure 10

A Selection CLB-CAg 9-90K+

B Selection IT-90K+

Deduced protein sequences of isolated FVIII A2 specific scFv

Heavy chains

```
              FR1                      CDR1      FR2              CDR2               FR3                                  CDR3           FR4
                                                                                                                                        1    1
              1         2         3              4         5         6         7         8         9         1         1               1    0    1
        12345678901234567890 12345 6789012345 6789012345 012a34567890012345 6789012345678901234 567890abcde12 34567890123
DP-10   QVQLVQSGAEVKKPGSSVKVSCKASGGTFS SYAIS WVRQAPGQGLEWMG GIIPIFGTANYAQKFQG RVTITADESTSTAYMELSSLRSEDTAVYYCAR ------EL DWFYI WGQGTMVTVSS
                    -------------------- --H-- -------------- D----L--G-------- ---------------T-T------------- ------T-T
DP-47   EVQLLESGGGLVQPGGSLRLSCAASGFTFS SYAMS WVRQAPGKGLEWVS AISGSGGSTYYADSVKG RFTISRDNSKNTLYLQMNSLRAEDTAVYYCAK RGRGGYKYYGMDV WGQGTTVTVSS
                    ----V----D----------- -NF-- ----------A--- --G-RS-T-P-------- ---------V-E--------I-
```

Figure 11A

Figure 11B

+1 Glu Val Gln Leu Val Glu Ser Gly Gly Asp Leu Val Gln Pro Gly Gly Ser
GAGGTGCAGC TGGTGGAGTC TGGGGGAGAC TTGGTACAGC CTGGGGGGTC 50
CTCCACGTCG ACCACCTCAG ACCCCCTCTG AACCATGTCG GACCCCCCAG

+1 Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Asn Phe Ala Met
CCTGAGACTC TCCTGTGCAG CCTCTGGATT CACCTTTAGC AACTTTGCCA 100
GGACTCTGAG AGGACACGTC GGAGACCTAA GTGGAAATCG TTGAAACGGT

+1 Ser Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val Ala Ala
TGAGCTGGGT CCGCCAGGCT CCCGGGAAGG GGCTGGAGTG GGTCGCGGCT 150
ACTCGACCCA GGCGGTCCGA GGGCCCTTCC CCGACCTCAC CCAGCGCCGA

+1 Ile Gly Gly Arg Ser Gly Thr Thr Phe Tyr Ala Asp Ser Val Lys Gly Arg
ATTGGCGGTA GAAGTGGTAC CACATTCTAC GCGGACTCCG TGAAGGGCCG 200
TAACCGCCAT CTTCACCATG GTGTAAGATG CGCCTGAGGC ACTTCCCGGC

+1 Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Val Tyr Leu Glu Met Asn
GTTCACCATC TCCAGAGACA ATTCCAAGAA CACGGTCTAT CTGGAAATGA 250
CAAGTGGTAG AGGTCTCTGT TAAGGTTCTT GTGCCAGATA GACCTTTACT

+1 Ser Leu Arg Ala Glu Asp Thr Ala Ile Tyr Tyr Cys Ala Lys Arg Gly
ACAGTCTGAG AGCCGAGGAC ACAGCCATTT ATTACTGTGC GAAAAGAGGG 300
TGTCAGACTC TCGGCTCCTG TGTCGGTAAA TAATGACACG CTTTTCTCCC

+1 Arg Gly Gly Tyr Lys Tyr Tyr Gly Met Asp Val Trp Gly Gln Gly Thr Thr
CGCGGGGGGT ATAAGTATTA TGGGATGGAC GTCTGGGGCC AGGGGACCAC 350
GCGCCCCCCA TATTCATAAT ACCCTACCTG CAGACCCCGG TCCCCTGGTG

+1 Val Thr Val Ser Ser
GGTCACCGTG TCGAGT
CCAGTGGCAC AGCTCA
400

Figure 11C